TITLE

"Soil amelioration by coal fly ash and its impact on growth and productive behaviour of *Withania somnifera* (Linn.) Dunal."

INTRODUCTION

In India about 75% electricity production is dependent on coal which produces an enormous quantity of fly ash as waste product from thermal power stations. About 70% of India's annual coal production is used in about 120 power plants to produce 112 million tones of coal fly ash per year and it may cross over 225 million tones up to 2017 (Kumar *et al.* 2005) and the current worldwide production of fly ash is more 700 million tons (Lokeshappa *et al.* 2011).

This fly ash is a resultant of combustion of coal at high temperature and regarded as a problematic solid waste all over world. Fly ash is disposed off either by dry or wet methods. In dry disposal, it is dumped in fly ash basin and landfills, while in wet method it is washed out with water into artificial lagoons called pond ash. Both of these methods finally leads to disposal of fly ash on an open area which degrades the soil and endangers environment as well as human health (Page *et al.*1979). Management of fly ash attained an apparent scenario for scientific and strategic concern due to large scale production of it (Ansari *et al.* 2011).

Physically fly ash occurs as a very fine particles of diameter <10 μm with light texture and high surface area. Chemical composition of fly ash depends on various factors such as parental coal, quality of coal used in power plants, operating conditions of thermal power station. 95 to 99% fly ash consist of oxides of Si, Al, Fe, Ca and about 0.5 to 3.5% consist of Na, P, K, S and the remainder of ash is composed of trace elements. Fly ash consists of all elements present in soil except organic carbon and nitrogen (Vijayan 2000).

Fly ash with these qualities has a vast potential to be used for amendment in agriculture. Its physico-chemical conditions are conductive for plant growth as macro and micronutrients are present in it. On the contrary fly ash also contains toxic trace elements but its effect on plant growth is not alarming and is found within the normal acceptable range. Hence, fly ash has got more beneficial qualities for plant growth and yield, as was investigated by (Elseewi *et al.*1978, Fisher *et al.* 1978, Adriano *et al.* 1980, Singh *et al.* 1994,

Khan *et al.* 1996, Khan *et al.* 1997, He *et al.* 1999, Vijayan *et al.* 1999, Gangloff *et al.* 2000, Grewal *et al.* 2001, Sharma *et al.* 2001).

Most of the work was done in crop plants to investigate the responses of plants to fly ash ameliorated soil and very few in medicinal plants therefore, *Withania somnifera* (Linn.) Dunal a potential drug yielding plant was selected for the present study. *Withania somnifera* (Linn.) Dunal also known as ashwagandha, Indian ginseng and winter cherry, belongs to family Solanaceae, an important herb in the ayurveda and indigenous medical system for over 3000 years. It possesses more potent antioxidant activity than common dietary plants (Li *et al.* 2007). Whole plant is used in preparation of medicine due to its usefulness as antioxidant, adaptogen, aphordisiac, liver tonic, anti inflammatory agent and astringent. Recently it is being used in treatment of ulcers, bacterial infection, venom toxins and senile dementia (Bhatnagar *et al.* 2005, Kumar *et al.* 2006, Machiah *et al.* 2006, Naidu *et al.* 2006). Clinical trial supports the use of ashwagandha for anxiety, cognitive and neurological disorders, inflammation, hyperlipidemia and Parkinsons disease (Gautam *et al.* 2004, Rasool *et al.* 2006). Ashwagandha's chemo preventive properties make it a potentially useful adjunct for patients undergoing radiation and chemotherapy (Ichikawa *et al.* 2006, Mathur *et al.* 2006). Recently Ashwagandha is also used to inhibit the development of tolerance and dependence on chronic use of various psychotropic drugs (Gupta *et al.* 2007).

In light of above investigations, present research work is planned to utilize the potential of fly ash in soil amelioration and impact of ameliorated soil on growth and reproductive behaviour of medicinal plant *Withania somnifera* (Linn.) Dunal.

A BRIEF REVIEW OF THE WORK ALREADY DONE IN THE FIELD

On the related topic many Indian as well as foreign reviews are covered which are as follows. Fly ash is not waste product but is beneficial in agriculture and horticulture purpose and can be used as soil amendment was proved in his study by Adriano *et al.* (1978). Fly ash act as macro and micronutrients provider for plants was studied by Vijayan *et al.* (1999). Fly ash can also help in restoring the ion balance in wastelands there by rendering them suitable for plant raising and can be used for reclamation of wasteland, agriculture, forestry, green house studies as it has capability of altering physical, chemical and microbial status of soil was investigated by many workers in India Singh *et al.* (1994), Khan *et al.* (1996), Karla *et al.* (1997), Kumar *et al.* (2000), Pathan *et al.* (2001), Gupta *et al.* (2002), Mishra *et al.* (2007),

Punjwani *et al.* (2011) and abroad by Adriano *et al.* (1980), Chang *et al.* (1989), Wong *et al.* (1989), Clark *et al.* (1995), He *et al.* (1999), Tuna *et al.* (2005).

Various studies have been done all over the world in few decades to determine the changes in properties of soil after addition of fly ash by many workers like Chang *et al.* (1977), Page *et al.* (1979), Adriano *et al.* (1980), Hill *et al.* (1980), Aitken *et al.* (1984), Gangloff *et al.* (2000) in abroad. Similar studies were done in India by Singh *et al.* (1994), Lal *et al.* ((1996), Khan *et al.* (1997), Sharma *et al.* (2001), Pathan *et al.* (2003), Gupta *et al.* (2007), Lokeshappa *et al*. (2011).

Change in physical properties like texture, bulk density, particle size distribution, porosity and water holding capacity in the soil ameliorated by fly ash was investigated by many workers in various parts of world like Adriano *et al.* (1980), Aitken *et al.* (1984), Brady *et al.* (1996), Gangloff *et al.* (2000), Zhang *et al.* (2004) in abroad. Similar studies was done by Singh *et al.* (1994), Arshed *et al.* (1996), Kumar *et al.* (2000), Gupta *et al.* (2006), Mishra *et al.* (2007) and Punjwani *et al.* (2011) in India.

Beneficial changes in chemical properties of soil ameliorated by fly ash like pH, electrical conductivity and elemental concentration were investigated by many researchers like Elseewi *et al.* (1978), Ciravolo *et al.* (1979), Elseewi *et al.* (1980), Aitken *et al.* (1984), El-Mogazi *et al.* (1988), Wong *et al.* (1989) and Cline *et al.* (2000) in abroad, similarly in India by Khan *et al.* (1997), Vijayan *et al.* (1999), Karla *et al.* (2003), Yunusa *et al.* (2006), Gupta *et al.* (2007), Mishra *et al.* (2007) and Punjwani *et al.* (2011).

Different levels of fly ash shows varied affects on seed physiology of various crop plants was studied by Wong *et al.* (1989), Sims *et al.* (1995) and Jaramillo-Lopez *et al.* (2011) in abroad and in India by Grewal *et al.* (2001), Karla *et al.* (2003), Mishra *et al.* (2007), Shrivastava *et al.* (2008) and Punjwani *et al.* (2011).

Major studies had been done to estimate germination percentage, growth, productive behaviour and yield of plants grown in fly ash ameliorated soil. Various crop plants like wheat, soyabean, rye, barley, rice, maize, tomato, til, mustard, clover, gram were grown in pot culture, green house and field by many workers like Page *et al.* (1977), Fox *et al.* (1990), Summer *et al.* (1998), Cline *et al.* (2000) and Tuna *et al.* (2005) in abroad similar studies was done in India by Lal *et al.* (1996), Khan *et al.* (1997), Singh *et al.* (1997), Sharma *et al.*

(2001), Rai *et al.* (2004), Garg *et al.* (2005), Mishra *et al.* (2005), Mishra *et al.* (2007) and Punjwani *et al.* (2011) and obtained beneficial results of fly ash on these plants.

Majority of investigations had been done all over the world for crop plants to find out the use of fly ash in soil amelioration and its effect on plant growth, productive beheviour and yield capacity but very few works had been done on medicinal plants. *Withania somnifera* (Linn.) Dunal known as ashwagandha of family Solanaceae, used as an antioxidant, adaptogen, aphrodisiac, liver tonic, anti-inflammatory agent, astringent and more recently found to be used in treatment of ulcers, bacterial infection, venom toxins and senile dementia. This was observed by Bhatnagar *et al.* (2005), Kumar *et al.* (2006), Machiah *et al.* (2006) and Naidu *et al.* (2006). The presence of biologically active chemical constituents like alkaloids (ashwagandhin, cuscohygrine, anahygrine, tropin, *etc*.), steroidal compounds, including ergostane type steroidallactones, withaferin A, withanolides A-Y, withasomniferin-A, withasomidienone, withasomniferols A-C, withanone, in Ashwagandha plant was investigated by Elsakka *et al.* (1990) and Ganzera *et al.* (2003).

OBJECTIVES

The present research proposal is designed to find out the feasibility of coal fly ash on amelioration of physico-chemical properties of soil and on growth responses, production behaviour of *Withania somnifera* (Linn.) Dunal under *invitro* culture. The study will be conducted to find out the following objectives:

1. To study the potential of coal fly ash to ameliorate the physical properties of soil.
2. To assess the way to enrich the chemical properties of soil by adding Fly ash.
3. To investigate an impact of fly ash on seed germination and growth responses.
4. To analyze the impact of Fly ash incorporation on medicinal value of *Withania somnifera* (Linn.) Dunal.
5. Evaluation of productive behaviour of *Withania somnifera* (Linn.) Dunal under pot culture.
6. To observe the yield characteristics of *Withania somnifera* (Linn.) Dunal under the influence of fly ash incorporated soil.
7. To evaluate the overall impact of coal fly ash on soil properties and growth responses, production behaviour of *Withania somnifera* (Linn.) Dunal by using statistical method.

NOTEWORTHY CONTRIBUTION IN THE FIELD OF PROPOSED WORK

Majority of work had been done to determine the impact of various concentration of fly ash incorporation on growth and productive behavior of several crops plants such as gram, rice, mustard, wheat, sorghum, tomato, spinach, soya bean, maize etc. but very little work has been done in medicinal plants. However, the noteworthy contributions have been made by the following workers which are Vijayan *et al.* (1999) had worked on the ability of fly ash to provide macro and micronutrients except organic carbon to plants. Physical properties of soil is altered by application of coal fly ash this fact was studied by various workers like Page *et al.* (1979), Aitken *et al.* (1984), Kumar *et al.* (2000), Mishra *et al.* (2007), Pujawani *et al.* (2011).

Change in chemical properties of soil was studied by Elseewi *et al.* (1978), Khan *et al.* (1996), Vijayan *et al.* (1999), Yunusa *et al.* (2006). Impact of fly ash on seed germination and productive behavior of plants like soyabean, corn, rice, tomato etc. was studied by Wong *et al.* (1989), Mishra *et al.* (1986), Khan *et al.* (1997), Cline *et al.* (2000), Jaramillo-Lopaz *et al.* (2011). Fly ash can be used as secondary source of nutrients on 10% and 20% concentration to increase soil fertility and biomass was investigated by Jala (2005). Impact of fly ash on soil properties, germination and production of Kalmegh (*Andrographis paniculata*) was investigated by Shrivastava *et al.* (2008).

PROPOSED METHODOLOGY

Plan of work

Pot culture experiments with three replicates will be conducted to analyze the feasibility of various levels (5% to 100%) of fly ash incorporation to soil on physico-chemical properties of soil, seed germination, growth responses, medicinal values and productive behaviour in *Withania somnifera* (Linn.) Dunal.

Physico-chemical analysis of soil sample

Physical and chemical properties of soil sample treated with fly ash will be determined.

- **Texture**

  Particle size distribution of fly ash treated soil sample will be assessed by following the method of Misra *et al.* (1970). Soil texture will be determined by soil texture triangle given by Palmer *et al.* (1977).

- **Hydrogen ion concentration (pH), electrical conductivity, total dissolved salts**

  Hydrogen ion concentration (pH), electrical conductivity, total dissolved salts of soil sample and fly ash will be determined by methods prescribed by American Public Health Association (APHA), American Water Works Association (AWWA) and Water Environment Federation (WEF) (1992).

- **Porosity (%) and soil solids (%)**

  By following the method of Misra *et al.* (1970) porosity and soil solids will be calculated in sample of various treatments.

- **Bulk density and particle density**

  Bulk density and particle density of soil sample treated with fly ash will be determined respectively by following the method of Blake *et al.* (1986).

- **Water holding capacity (WHC)**

  Water holding capacity (WHC) will be estimated by the following the method of Pandeya *et al.* (1968).

<u>Growth responses, medicinal values and productive behavioral studies</u>

In present investigation seed germination, growth responses, medicinal values and productive behaviour of *Wathania somnifera* (Linn.) Dunal will be determined by estimating following characteristics:

Characteristics of seeds

- **Moisture content**

  The moisture content of seeds will be estimated in percentage (%) by using the formula:

$$\text{Moisture content (\%)} = \frac{\text{(Fresh weight – Dry weight)}}{\text{(Fresh weight)}} \times 100$$

- **Rate of germination**

  Rate of germination will be assayed in term of percent germination by using the following formula:

$$\text{\% Germination} = \frac{\text{Total number of seeds kept for germination}}{\text{Number of germinated seeds}}$$

- **Mean daily germination (MDG)**

  Mean daily germination will be estimated by using the following formula:

$$\text{Mean daily germination} = \frac{\text{Total number of seeds germinated}}{\text{Days/Time required for germination}}$$

- **Peak value (PV)**

  Peak value will be estimated by using the formula:

$$\text{Peak value} = \frac{\text{Maximum germination on a day}}{\text{Days/Time taken to achieve it}}$$

- **Germination index (GI)**

  Germination index will be determined by using the formula:

  Germination Index = Mean daily germination x Peak value

- **Average seedling length (in cm)**

  Length of seedling will be measured by Vernier calipers.

- **Average seedling dry weight (in gm)**

  Dry weight of seedling will be measured by using electronic balance.

- **Vigour index (VI)**

  Vigour index of seeds in various soil treatments would be calculated by using following formula:

  Vigour index = % Seed germination x Seedling dry weight

Growth responses in *invitro* pot culture experiments

In pot culture experiments cow dung/vermicompost will be used as source of organic matter and nitrogen uniformly in pots of various fly ash treatments. Seeds will be potted after surface sterilization. In this % germination will be estimated and plant will be observed for following morphological characters and productive behaviour.

- **Root length, shoot length, number of leaves per plant, leaf area per leaf, leaf area per plant, number of branches**

  Root length, shoot length, number of leaves per plant, leaf area per leaf, leaf area per plant, number of branches will be measured.

- **Chlorophyll a, Chlorophyll b, Carotenoid contents**

  Amount of chlorophylls and carotenoid will be calculated by following the method of Arnon (1949) and Duxbury *et al.* (1956) and the pigment concentration will be expressed in mg per gm dry matter.

- **Dry biomass of root, shoot and plant**

  Weight will be taken by using electronic balance. Dry biomass of root, shoot and plant will be assessed after drying them separately in oven at 105 ºC for 24 hrs.

- **Moisture content (%) of plant material**

  Percent of moisture content of root, shoot and plant will be calculated by following formula:

$$\text{Moisture content (\%)} = \frac{\text{(Fresh weight – Dry weight)}}{\text{(Fresh weight)}} \text{ X } 100$$

Medicinal values

Flavonoid, total phenol and total alkaloid will be measured by standard methods.

Functional aspects of plant production

To find out the functional aspects of plant production following will be measured:

- **Root shoot ratio**, **root weight ratio, shoot weight ratio**

  Root shoot ratio, root weight ratio, shoot weight ratio will be measured to observe the impact of fly ash treatment.

- **Leaf area ratio (LAR)**

  Leaf area ratio will be assessed by using the method of Grantz *et al.* (2006)**.**

- **Mean leaf : weight ratio (MLWR)**

  Mean leaf : weight ratio will be calculated by following the method of Blackman (1968).

- **Growth rate**

  It will be calculated by using following formula and expressed in gm day^{-1}.

$$\text{Growth rate} = \frac{(W_2-W_1)}{(T_2-T_1)}$$

  Where; W_1 and W_2 are dry weight of plant at time T_1 and T_2.

- **Relative growth rate:**

  Relative growth will be estimated by the method of Blackman (1968).

Characteristics of harvested seeds

Average number of fruit per plant, number of seeds per fruit, seed output per plant, seed yield per plant (in gm), 1000 grain weight (in gm) will be determined and average seed germination (%), average seedling length (in cm), average dry weight of seedling (in gm), vigour index and reproductive capacity of harvested seeds will be calculated by using standard methods.

Statistical analysis

Statistical analysis will be done to find out the significant correlations amongst the data collected from experiments.

EXPECTED OUTCOME OF THE PROPOSED WORK

1. The fly ash management could be done by using fly ash in soil amelioration.

2. The potential of fly ash could be determined in soil amelioration.

3. Quantity of fly ash required for soil amelioration could be determined.

4. Soil quality could be determined before and after the amelioration by fly ash.

5. Growth and productive response of medicinal plant *Withania somnifera* (Linn.) Dunal in ameliorated soil could be known.

6. Medicinal value of *Withania somnifera* (Linn.) Dunal in ameliorated and non ameliorated soil could be assessed.

7. Responses of *Withania somnifera* (Linn.) Dunal to ameliorated soil could be compared with the crop plants, as most of the work has been done by the workers on crop plant.

8. The outcome of the proposed work will be helpful in planning fly ash management strategy.

9. Suggestion could be made to cultivators for the utilization of Fly ash as a cheaper and potent source of nutrients.

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Kumar, T., Sahu, K.K. and Naithani, S.C. 2010. Antioxidants of healing herbs: *Withania* and *Rauwolfia*. Lambert Academic Publishing AG & Co. KG, Germany.

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